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# Monitoring Sensitive Bat Species at Los Alamos National Laboratory

By

Kari M. Schoenberg

## Abstract

Bats play a critical role in ecosystems and are vulnerable to disturbance and disruption by human activities. In recent decades, bat populations in the United States and elsewhere have decreased tremendously. There are 47 different species of bat in the United States and 28 of these occur in New Mexico with 15 different species documented at the Los Alamos National Laboratory (LANL) and surrounding areas. *Euderma maculatum* (the spotted bat) is listed as “threatened” by the state of New Mexico and is known to occur at LANL. Four other species of bats are listed as “sensitive” and also occur here. In 1995, a four year study was initiated at LANL to assess the status of bat species of concern, elucidate distribution and relative abundance, and obtain information on roosting sites. There have been no definitive studies since then. Biologists in the Environmental Protection Division at LANL initiated a multi-year monitoring program for bats in May 2013 to implement the Biological Resources Management Plan. The objective of this ongoing study is to monitor bat species diversity and seasonal activity over time at LANL. Bat species diversity and seasonal activity were measured using an acoustic bat detector, the Pettersson D500X. This ultrasound recording unit is intended for long-term, unattended recording of bat and other high frequency animal calls. During 2013, the detector was deployed at two locations around LANL. Study sites were selected based on proximity to water where bats may be foraging. Recorded bat calls were analyzed using Sonobat, software that can help determine specific species of bat through their calls. A list of bat species at the two sites was developed and compared to lists from previous studies. Species diversity and seasonal activity, measured as the number of call sequences recorded each month, were compared between sites and among months. A total of 17,923 bat calls were recorded representing 15 species. Results indicate that there is a statistically significant relationship between bat diversity and month of the year. Future studies will be implemented based on these findings.

## Introduction

There are more than 1,300 species of bats worldwide (Harvey et al. 2011). That's about one-fifth of all mammal species known today. In fact bats are the second largest group of mammals in the world (rodents being the first with 2,200+ species) (Harvey et al. 2011). Except for the polar-regions and remote islands (Jones et al. 2009), bats live in almost every habitat on Earth. Bats play critical roles in ecosystem function. They consume enormous quantities of agricultural and forest pests and reduce the need for pesticides (O'Shea et al. 2003). Some are critical pollinators and seed-dispersers for plants, many with great economic value (Harvey et al. 2011; Jones et al. 2009). However, bat populations can be very vulnerable to disturbance and disruption by human activities and many are threatened worldwide. Bats are among the least studied and most misunderstood of animals. In recent decades, bat populations in the United States and elsewhere have decreased tremendously (Bogan et al. 1997). White-nose syndrome, discovered in the northeastern United States, is responsible for well over a million of these die-offs (Harvey et al. 2011). It is caused by a white fungus that grows on the muzzles, ears and wing membranes of affected bats (Jones et al. 2009).

Bats belong to the Order Chiroptera meaning “hand wing” (Harvey et al. 2011). There are two types of bat in the world, Megachiroptera, which include fruit eating bats, and Microchiroptera, which eat mostly insects (Harvey et al. 2011; Huston et al. 2001). Most bats can eat up to 50% of their body weight in a single night (Harvey et al. 2011). Bats do not like the cold and so many will migrate or hibernate in the winter. For their size bats are the slowest reproducing mammals on Earth. They give birth once a year and typically produce only one pup (Harvey et al. 2011). Gestation will last up to four weeks and babies are typically born in May or June. When they are born, pups can weigh up to 40% of their mother's body weight (Harvey et al. 2011).

There are 47 different species of bats in the United States (Harvey et al. 2011) and 28 of these occur in New Mexico with 15 different species documented at the Los Alamos National Laboratory (LANL) and surrounding areas (Bogan, et al. 1997). In 1995, a four year study was initiated at LANL to assess the status of bat species of concern, elucidate distribution and relative abundance, and obtain information on roosting sites. There have been no definitive studies since then.

Biologists in the Environmental Protection Division at LANL initiated a preliminary study for a multi-year monitoring program for bats in May 2013 to implement the Biological Resources Management Plan. The objective of this on-going study is to monitor bat abundance and species richness over time at LANL. Specific line of inquiry include *how does the current list of bat species compare to historical data; do some sites at LANL receive more use by bats than other sites; and how has habitat change/climate change affected the bat community?* Another objective is to look for sensitive bat species that use LANL sites. *Euderma maculatum* (the spotted bat) is listed as “threatened” by the state of New Mexico and is known to occur at LANL ([BISON-M]

Biota Information System of New Mexico). Four other species of bat considered “sensitive” also occur at LANL. They are *Corynorhinus townsendii* (the Townsend’s big-eared bat), *Myotis ciliolabrum* (the small-footed bat), *Myotis volans* (the long-legged bat), and *Nyctinomops macrotis* (the big free-tailed bat) (Bogan, et al. 1998; [BISON-M] Biota Information System of New Mexico).

## Methods

### Study Locations

A bat detector (discussed in the next section) was deployed at two locations within the LANL site to determine use of the sites by bat species. Study sites were selected based on proximity to permanent water where bats may be foraging. The first site was located on Sigma Mesa (Figure 1) at the North end of the evaporation ponds. The second site was located at the head of the Sandia Wetlands (Figure 2).



**Figure 1.** Sigma Mesa at the North end of the evaporation ponds. The microphone for the detector is pointed towards the water.





**Figure 2.** At the head of the Sandia Wetlands.

## Data Collection

Most Microchiroptera use echolocation to navigate around obstacles and capture prey in the dark. These sounds are mostly emitted in the high frequency (above 20 kHz) range and are above the hearing range of humans (Hill & Smith 1984). Bat species abundance and richness was measured using an acoustic bat detector, the Pettersson D500X (Figure 3). The D500X is an ultrasound recording unit intended for long-term, unattended recording of bat calls. It records full-spectrum virtually no gaps system allows the recording as a sound is

Two removable 16 GB for data storage. The passively record data via an internal clock on in a waterproof housing precipitation. The frequency of 500 kHz and



**Figure 3.** Pettersson D500X

ultrasound in real time with between recordings. The trigger device to automatically start detected.

compact flash cards were used detector was programmed to between 7:00 pm and 7:00 am the detector and was deployed to give protection from detector was set with a sampling trigger sensitivity low. The trigger

sensitivity setting determines how long a signal must be before triggering a recording. It was set to low in order to reduce ambient noise files without losing bat calls. The rest of the recording settings were as follows; INPUT GAIN=80, TRIG LEV=80, INTERVAL=0, MODE=AUTO, TIMEOUT=05, PRETRIG=OFF, REC.LEN=5, HP-FILTER=OFF, AUTOREC=YES. An explanation of what each setting is for is discussed in more detail in Appendix 1.

The detector was checked about once a week to determine functionality and to collect the digital data. Once full, cards were collected and displayed on a computer using a card reader. Software from the Pettersson website was used to read and archive the data recorded. Older detectors such as Anabat used in previous bat studies at LANL had to be manually compared to reference libraries in order to determine bat species. This newer system could be more systematic for species identification.

## **Analysis of Calls**

Call files were extracted from data files using Sonobat software (Figure 4). Sonobat software provides a comprehensive tool for analyzing and comparing high-resolution full-spectrum sonograms of bat echolocation calls recorded from full-spectrum and time-expansion bat detectors. The software performs a spectral analysis of the data to extract six dozen parameters that describe the time-frequency and time-amplitude trends of a bat call. Sonobat then uses a decision engine based on the quantitative analysis of approximately 10,000 species-known recordings from across North America and compares the data with the separate files. In Sonobat ultrasound recordings of bat echolocation may be broken into recordings of a single bat call or recordings of bat call sequences to extract call parameters and to classify to species. A call is a single pulse of sound produced by a bat, while a call sequence is a combination of two or more pulses recorded in a file. Classifying an entire sequence will typically provide more reliable results as this method benefits from the combined information within the sequence. During the Sonobat batch run species names were added to the end of the file name. After the Sonobat batch run the output can be saved to an excel spreadsheet where data was analyzed to determine what species of bats were recorded at each specific site. Many calls that were unable to be identified were most likely triggered by insects above 25 kHz or the call was too faint or below the quality that was identifiable by the software.



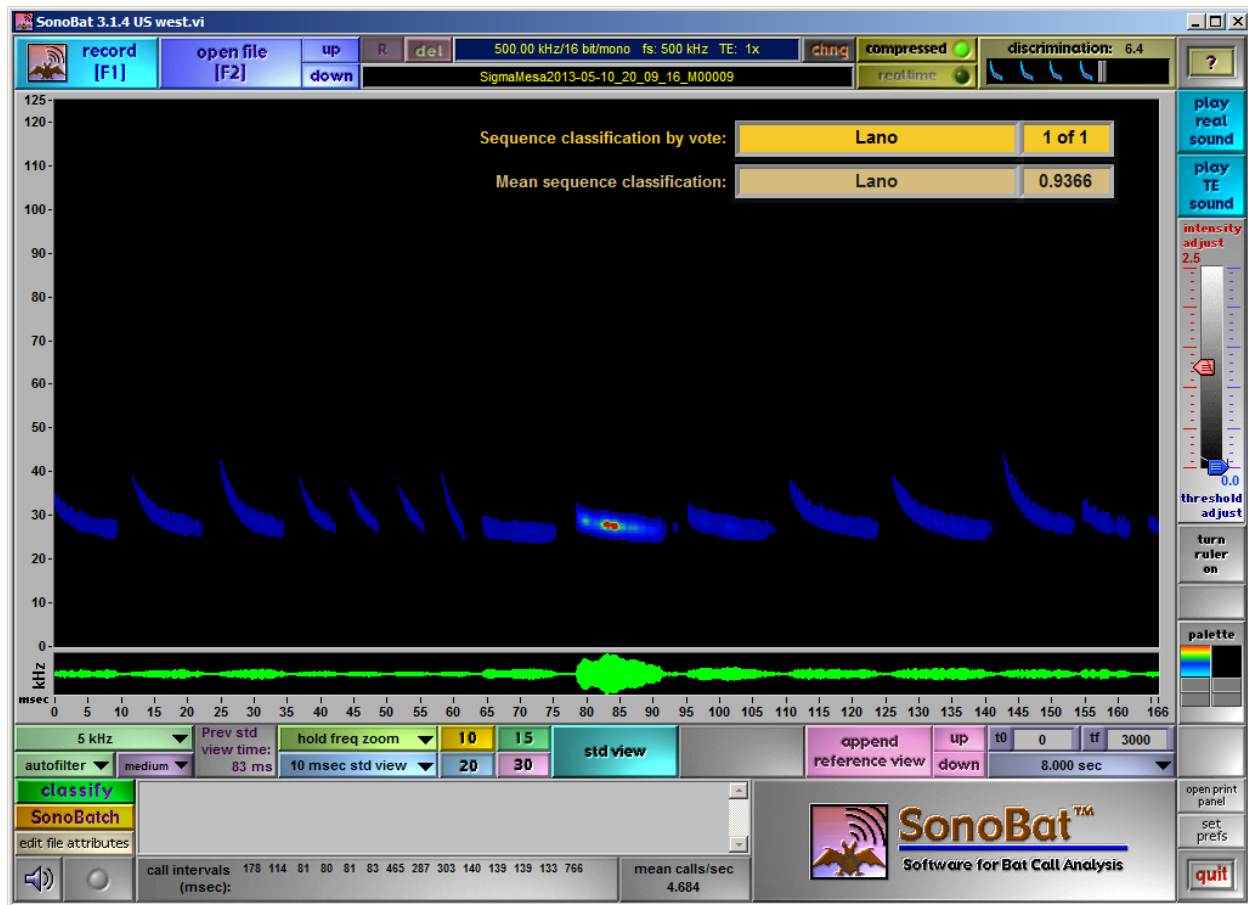


Figure 4. Screen shot of the Sonobat analysis. The blue lines indicate the audible frequency of bat calls recorded. Sonobat will classify individual calls or an entire sequence and label it in the top right corner which is shown here in the yellow boxes.

## Statistical Methods

The Shannon's diversity index ( $H$ ) (Shannon 1948) was used to compare species diversity between sites and among month. This is a popular diversity index in ecology used to describe the species richness in a community. The Shannon's  $H$  can range from 0.0 to 4.6, where larger values represent increasing diversity.  $H$  is calculated using the following formula:

$$H = -1(p_i (\ln(p_i)))$$

Where  $p_i$  is a percentage value of a specific species in the total population and  $\ln$  is the natural log.

In order to compare indices a bootstrapping technique is used. This is a resampling procedure for estimating the distributions of statistics based on independent observations. Here two samples, A

and B, are pooled. Then random samples (of the same size) are drawn a large number of times (about 1,000) from this pool. The diversity indices and the bootstrap comparisons between indices were completed using free statistical software called PAST (PAleontological STatistics) (Hammer et al. 2001).

## Results

This was a preliminary study to determine the best course of action in starting a full multi-year study of bat species in the area. Based on analysis of bat calls the following results were gained.

### Seasonal Activity

The acoustic detector was deployed on May 10, 2013 and continued to record data through September 30, 2013 for a total survey period of 145 calendar nights. The range dates that the detector was deployed at each location is summarized in Table 1. Equipment malfunction (dead batteries) caused some gaps in the data. Fire restrictions in late June caused data gaps as well while the detector was at the Sandia Wetlands. Due to these gaps it was decided to deploy the detector at the Sandia Wetlands for longer than at Sigma Mesa.

**Table 1. The dates the detector was deployed at each site.**

Location	Dates Deployed
Sigma Mesa	May 10 - June 25
Sandia Wetlands	June 25 - September 30
Fire restrictions put into place from June 25 - July 8	

Seasonal activity, measured as the number of call sequences recorded each month, varied at each location. A total of 17,928 call files were recorded by the detector at the two sites. Sigma Mesa had a total of 8,838 bat calls representing 15 species while the Sandia Wetlands had a total of 9,090 bat calls representing 14 species.

*Eumops perotis* (the western mastiff bat) was one species detected at Sigma Mesa but it is not clear whether or not this was an actual detection or a mistaken identification from the Sonobat software. *E. perotis* ranges from central Mexico across the southwestern United States (parts of

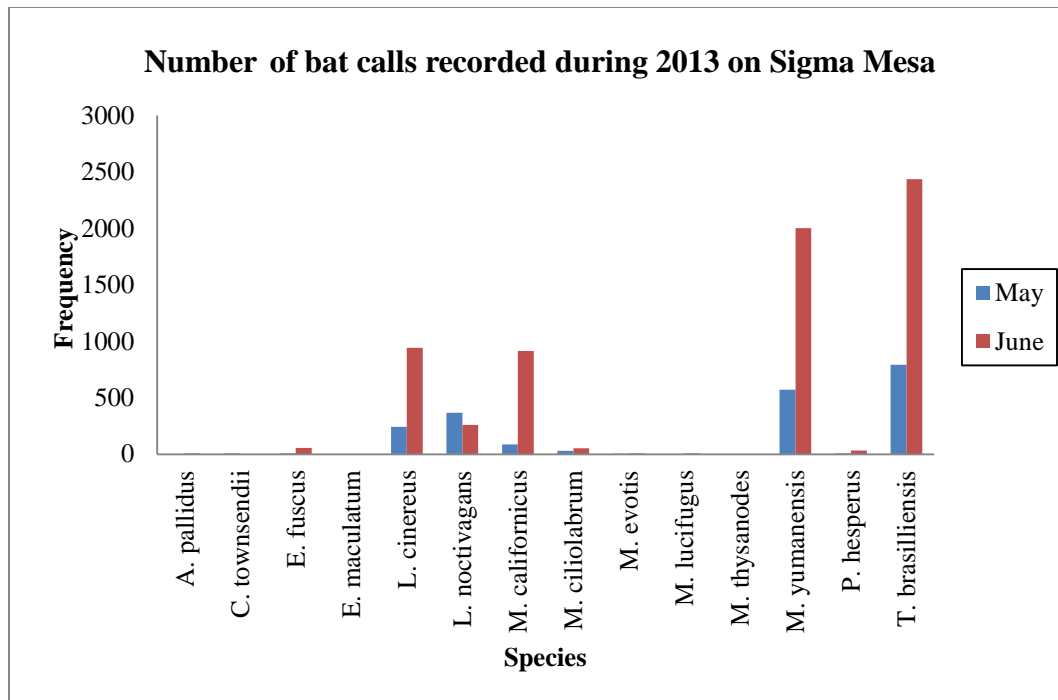
California, southern Nevada, Arizona, southern New Mexico and western Texas). Although it can enter a daily torpor, *E. perotis* does not hibernate and may be active year round. They do not appear to migrate either. While it can be found in New Mexico farther south it has never been documented in Los Alamos or in New Mexico this far north. Due to this, these recordings were considered outliers and were removed from the data set (Table 2). A total of 5 call files were identified as *E. perotis*. With *E. perotis* removed there were a total of 17,923 call files recorded from the two sites. Sigma Mesa then had a total of 8,833 bat calls representing 14 species. Due to this mistaken identification it was necessary to check with other data sources in order to verify the software was making sense.

**Table 2. List of numbers of detections of different bat species at Sigma Mesa during the study.**

Species	May	June	Total
<i>E. maculatum</i>	0	1	1
<i>M. lucifugus</i>	0	6	6
<i>A. pallidus</i>	2	6	8
<i>M. thysanodes</i>	2	0	2
<i>M. evotis</i>	3	8	11
<i>C. townsendii</i>	7	2	9
<i>P. hesperus</i>	7	33	40
<i>E. fuscus</i>	8	54	62
<i>M. ciliolabrum</i>	29	53	82
<i>M. californicus</i>	86	913	999
<i>L. cinereus</i>	242	943	1185
<i>L. noctivagans</i>	367	259	626
<i>M. yumanensis</i>	571	2002	2573
<i>T. brasiliensis</i>	793	2436	3229

*Myotis lucifugus* (the little brown bat) has not been documented in Los Alamos before either. However, it is more likely this species is found here as its range is a large portion of North America from Alaska south into central Mexico and is found all over New Mexico. It was also stated in previous studies that *M. lucifugus* is known from elsewhere in northern New Mexico and may occur in the Jemez Mountains, at least at lower elevations (Bogan et al. 1998). The software identified 158 calls as *M. lucifugus* at the two location sites and so it was decided not to remove this data.

Approximately three-fourths (75.3%) of the total calls were recorded during the month of June, when detection rates peaked at this location (Figure 5).



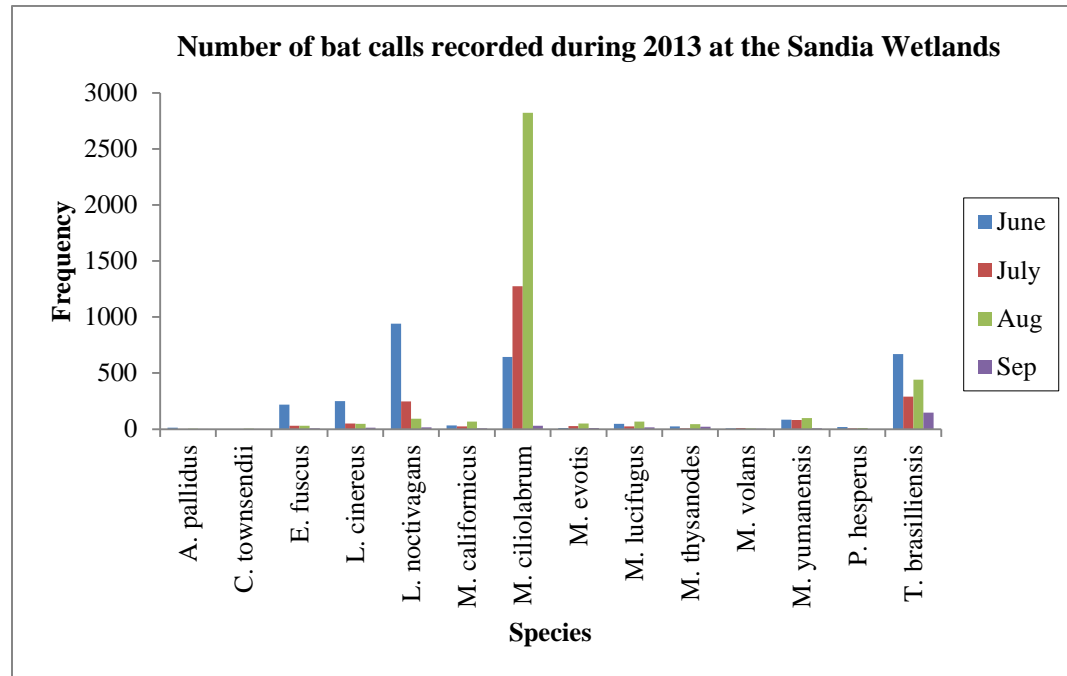
**Figure 5. Number of bat calls recorded in 2013 on Sigma Mesa.**

As mentioned before, the Sandia Wetlands had a total of 9,090 bat calls representing 14 species (Table 3). Approximately 31% of the total calls were recorded during the month of June with the plurality of the calls (41%) recorded during the month of August (Figure 6). Some of the variations could have correlated with reproduction and the young beginning to forage for themselves. Female bats usually give birth in June and feed pups on their milk. At six weeks old the young bats begin to catch insects for themselves and no longer need their mother's milk to survive.

**Table 3. List of numbers of detections of different bat species at the Sandia Wetlands during the study.**

Species	June	July	Aug	Sep	Total
<i>C. townsendii</i>	0	0	4	2	6
<i>M. volans</i>	6	11	8	3	28
<i>M. evotis</i>	10	26	49	10	95
<i>A. pallidus</i>	12	2	5	1	20
<i>P. hesperus</i>	18	6	10	0	34
<i>M. thysanodes</i>	24	8	45	21	98
<i>M. californicus</i>	34	23	67	6	130
<i>M. lucifugus</i>	46	25	65	15	151
<i>M. yumanensis</i>	84	80	98	6	268

<i>E. fuscus</i>	218	29	31	7	285
<i>L. cinereus</i>	250	49	47	12	358
<i>M. ciliolabrum</i>	644	1274	2824	30	4772
<i>T. brasillensis</i>	670	290	442	147	1549
<i>L. noctivagans</i>	942	247	92	15	1296



**Figure 6. Number of bat calls recorded in 2013 in the Sandia Wetlands.**

At the Sandia Wetlands location, the detector recorded more call sequences than the Sigma Mesa location. However this is mostly likely due to the fact that the detector was deployed for a longer period of time in the Sandia Wetlands to account for lapses in detector function.

## Species Composition

Species composition did not differ much at each location. At Sigma Mesa, the most frequently recorded species was *Tadarida brasiliensis* (the Mexican free-tailed bat), followed by *Myotis yumanensis* (the Yuma bat), *Lasiurus cinereus* (the hoary bat), *M. californicus* (the California bat), and *Lasiurus noctivagans* (the silver-haired bat). Other species recorded going from most frequent to least were *M. ciliolabrum*, *Eptesicus fuscus* (the big brown bat), *Parastrellus*

*hesperus* (the western Pipistrelle bat), *M. evotis* (the long-eared Myotis bat), *Corynorhinus townsendii*, *Antrozous pallidus* (the pallid bat), *M. lucifugus* (the little brown bat), *M. thysanodes* (the fringed Myotis bat), and *E. maculatum*. The most frequently recorded species at the Sandia Wetlands was *M. ciliolabrum*, followed by *T. brasiliensis*, *Lasionycteris noctivagans*, *Lasiurus cinereus*, and *E. fuscus*. Other species recorded going from most frequent to least were *M. yumanensis*, *M. lucifugus*, *M. californicus*, *M. thysanodes*, *M. evotis*, *Parastrellus hesperus*, *M. volans*, and *Antrozous pallidus*. By September bat activity had decreased significantly. Most all bat species detected at Sigma Mesa were also detected in the Sandia Wetlands. *E. maculatum* was the one species that was found at Sigma Mesa but not in the Sandia Wetlands and *M. volans* was the one species that was detected in the Sandia Wetlands but not at Sigma Mesa.

## Species Diversity

For Sigma Mesa June had the highest number of bat call recordings, while May had the highest species diversity (Table 5). At Sigma Mesa species diversity during May and June were found to not be statistically different from each other ( $p=0.472$ ).

**Table 5. Shannon diversity index for each month at Sigma Mesa. Indices with the same letter following the value are not significantly different from one another.**

Month	Diversity index
May	1.54a
June	1.53a
All surveys	1.56

The Sandia Wetlands saw the most bat call recordings in August, while June had the highest species diversity (Table 6). Here June was found to be statistically different ( $p=0.001$ ) than July and August while July was found to be statistically different than August and September ( $p=0.001$ ) and August was found to be statistically different than September ( $p=0.001$ ). June and September were not statistically different from each other ( $p=0.343$ ).

**Table 6. Shannon diversity index for each month at the Sandia Wetlands. Indices with the same letter following the value are not significantly different from one another.**

Month	Diversity index
June	1.78a
July	1.33b



August	1.04c
September	1.71a
All surveys	1.54

While both sites were compared to each other, it should be noted that the detector was deployed at the Sigma Mesa site for much less time compared to the Sandia Wetlands, and the detector was deployed at the sites sequentially, not simultaneously. Sigma Mesa had a statistically higher species diversity compared to the Sandia Wetlands ( $p=0.047$ ) (Table 7). Due to the differences in deployed dates, any significant differences between the two sites should be viewed with caution, especially since the greatest Shannon diversity index values for the Sandia Wetlands were higher than those recorded at Sigma Mesa.

**Table 7. Species diversity comparison for both sites. Indices with the same letter following the value are not significantly different from one another.**

Site comparison	Diversity index
Sigma Mesa	1.57a
Sandia Wetlands	1.54b
Both sites	1.85

## Discussion

A content analysis found that many of the same bat species detected during the study were found in the 1995-1999 study. A total of 15 different bat species were recorded in 2013 compared to 15 species found in 1995-99 (Table 8). The species *Nyctinomops macrotis* was documented in 1995, however was not recorded in this study. The 1995 study covered a larger range and noted that *Nyctinomops macrotis* was found at higher elevations in the Jemez Mountains. This species has also been documented roosting in Frijoles Canyon so there might be a geographic limitation to the single point acoustic sampling. This species is listed as “sensitive” in the state of New Mexico ([BISON-M] Biota Information System of New Mexico).

**Table 8. Comparison of bat species found in 1995-99 vs. 2013.**

Species	1995-99	2013
<i>Antrozous pallidus</i>	X	X
<i>Corynorhinus townsendii</i>	X	X
<i>Eptesicus fuscus</i>	X	X

Euderma maculatum	X	X
Lasiurus cinereus	X	X
Lasionycteris noctivagans	X	X
Myotis californicus	X	X
Myotis ciliolabrum	X	X
Myotis evotis	X	X
Myotis lucifugus		X
Myotis thysanodes	X	X
Myotis volans	X	X
Myotis yumanensis	X	X
Nyctinomops macrotis	X	
Parastrellus hesperus	X	X
Tadarida brasiliensis	X	X

Sigma Mesa had larger species diversity during the period monitored. This could be due to the fact that the water in the evaporation pond is stagnant and may have more bug activity. In past studies there were greater areas with water where bats were likely to forage. The years after have seen a decrease in water sources due to a drying climate change and therefor alteration in habitat. Not only has there been a decrease in water sources but a decrease in precipitation as well. This change could have caused many bats to change locations. This could also be the reason bat species diversity numbers are so low.

One *E. maculatum* was recorded on June 7 at Sigma Mesa. This species is listed as “threatened” in the state of New Mexico by the New Mexico Department of Game and Fish in 1988 ([BISON-M] Biota Information System of New Mexico). *E. maculatum* is distributed from south-central British Columbia to southern Mexico. It has been listed as threatened because of limited information available and uncertainty as to life history and population trends. These bats have been found in a variety of habitats but are more likely to occur in forested environments. *E. maculatum* roost in small crevices of cliffs and forage a regular circuit along with clearings in pine forests for prey (Harvey et al. 2011). Recorded species listed as “sensitive” by the state of New Mexico included *Corynorhinus townsendii*, *Myotis ciliolabrum*, and *Myotis volans* ([BISON-M] Biota Information System of New Mexico).

## Management Recommendations

A multi-year study with a larger study area will provide a long-term data set of the status and trends of bat populations around LANL and surrounding areas. A longer study will also help to

watch for sensitive species that use habitat located on LANL land. One test that might be done is to test for white nose syndrome. This is a fungal growth that infects cave roosting bats. It causes bats to rouse too frequently from hibernation. This results in a high rate of weight loss and bats starve to death before their food supply becomes available in the spring (Harvey et al. 2011). It was discovered in a cave in New York in 2006 and has spread rapidly across the eastern United States. Since its discovery over a million bats have died. White nose syndrome has not yet been documented in New Mexico but it might be beneficial to start testing for it in case it spreads into the southwest.

In previous studies suggestions were made to conduct a preliminary survey evaluating the potential impact of environmental contaminants on bats of this region. Samples taken at LANL sent in for analysis found no detectable contaminant levels but a targeted sampling in a known area of contamination would be a valuable study. Some contaminants have been documented as playing a role in some declining populations of bats in the United States. Bats could be exposed to some of these contaminants through drinking contaminated water at local sources on LANL, as well as from pockets of terrestrial contamination.

If there is a need to compare bat diversity between sites, at least two detectors should be used to allow simultaneous monitoring of sites. The results from this study suggest that monitoring in the late spring and early summer may be most effective at detecting the widest variety of bats.

## **Acknowledgements**

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## Appendix 1. Instrumentation

Internal batteries were used to power the Pettersson D500X detector. However, these batteries only lasted 3-4 days max before they had to be changed. An external battery was then purchased for the detector. The external battery lasted about two weeks at a time before needing to be recharged. While the external battery was being recharged I switched back to the internal batteries.

As stated in the methods recording settings for the detector included frequency, trigger sensitivity, input gain, trigger level, interval, mode, timeout, pre-trigger, rec. length, HP-filter, and autorec.

The trigger sensitivity setting determines how long a signal must be before triggering a recording. A “very high” setting will initiate a recording at the shortest signal duration. It was set to low in order to reduce ambient noise files without losing bat calls. The input gain is essentially the volume and should be adjusted to keep the recording level below the limit. The trigger level is adjusted to make the detector start recording at the desired level and the interval setting is just the minimal time interval to the next recording. Using a value greater than 0 means that at least the chosen time interval will have to elapse before the next recording is made. Mode and timeout are for display settings and can remain conservative for passive recordings. The Mode parameter determines in which situations the display is on. When set to Auto the display is turned off while the detector is asleep. The timeout parameter determines the time that the display remains on after being temporarily turned on with the ON/OFF key while the detector is in sleep mode. Pre-trigger determines how long the detector should start recording before the next sound. Using the detector without pre-trig (“PRE=OFF”, the recording starts as a sound triggers it) makes the detector use much less power while waiting for a sound. Recording length is how long the detector will record for once a sound has triggered it and the high pass (HP) filter attenuates low-frequency signals. To record signals with a frequency below ca. 15 kHz, the filter should be set to OFF. The detector can be set to either auto record or manual. For unattended recordings this setting should be set to automatic.